(*NEW SERIES.*)

No. 24.

SCIENTIFIC MEMOIRS

BY

OFFICERS OF THE MEDICAL AND SANITARY DEPARTMENTS

OF THE

GOVERNMENT OF INDIA.

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BY

CAPTAIN W. S. PATTON, M.B., I.M.S.

ISSUED UNDER THE AUTHORITY OF THE GOVERNMENT OF INDIA
BY THE SANITARY COMMISSIONER WITH THE GOVERNMENT
OF INDIA, SIMLA.

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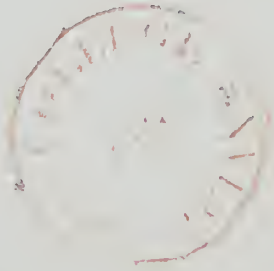

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ON A PARASITE FOUND IN THE WHITE CORPUSCLES OF THE BLOOD OF PALM SQUIRRELS.

SEVERAL *Hæmogregarinidæ* have been discovered recently in the blood of mammals, and in view of the great importance which attaches to such forms, the description of another example is of interest, especially as the parasite of this class which I have found, invades a special cell, the large mononuclear leucocyte.

The squirrel which harbours this parasite of the white blood corpuscles is the common, striped, palm squirrel of Kathiawar and Gujarat. It would appear on comparing this squirrel with the corresponding one in Madras, that although they resemble one another superficially they are distinct species. Brisson, in 1762, established the form with three pale dorsal stripes and a rufous band under the tail as being the typical *palmarum* L, and the Madras squirrel belongs to this species. The Kathiawar squirrel has five pale stripes and the rufous band under the tail is absent. Mr. Wroughton [1] has studied the two species and decided to separate them, naming the three striped species *Funambulus palmarum* and the five-striped form *Funambulus pennantii*, raising the latter to a new species.

From the results of examinations of the blood of 18 Madras squirrels, I think it is probable that the parasite which I propose to describe in this paper is confined to the Kathiawar species.

Parasites belonging to the genus *Hæmogregarina* have, until quite recently, been found only in cold-blooded animals, namely, *Batrachia*, *Chelonia*, *Crocodilia*, *Lacertilia* and *Ophidia*.

The parasites have the following characters: they are elongated, fusiform bodies with the appearance of vermicules and are seen lying coiled up in the cells or free in the plasma. They do not produce pigment and have no sexual form of development. In cold-blooded animals, they are confined entirely to the red cells.

The first was found by Lankester in the blood of *Rana esculenta* and described by him in 1871; since then no less than 19 species have been described. Quite recently two more have been described in frogs, *H. neireti* [2] and *H. theileri*,[2] one in an African toad,[3] one in a lizard, *H. varani*,[2] and one in a tortoise, *H. testudinis*.[2]

It was universally believed, that *Hæmatozoa* of this genus were confined to cold-blooded animals. It is now known that this is not the case, as they have been found in the blood of mammals.

Apparently the first mammalian hæmogregarine discovered was a parasite found by Dr. Bentley in 1904 in the white blood corpuscles of a dog in Assam. This parasite has been described by Captain James, I.M.S.[4] It is seen in the leucocytes and more particularly in the polymorphonuclear variety.

It appears as an elongated bean-shaped body composed of very slightly granular protoplasm and lies in the substance of the polymorphonuclear leucocytes. The nucleus is a large, oval or round chromatin mass. Each body consists of a definite cytocyst and a parasite enclosed within it. It contains no pigment granules. The leucocytes containing these bodies show no degenerative change, and they are not less amæboid than the unaffected leucocytes.

James describes two forms, one in which the protoplasm stains uniformly blue and contains, with the exception of the nucleus, no chromatin granules; the other in addition to the nucleus contained a definite faintly-stained centrosome situated near one or other side of the parasite and surrounded by a circular area of unstained protoplasm. A few chromatin granules may be seen scattered throughout the substance of the parasite.

He describes one of the parasites found free in the plasma in a stained film. It was almond-shaped but thicker at one end than the other. The nucleus was elongated and central and the centrosome was distinctly seen. In two instances he observed two parasites lying in one leucocyte.

In fresh preparations the parasite appeared as a refractile elongated body lying in the protoplasm of the polymorphonuclear leucocytes. One of the parasites was observed to escape from a cell; it performed slow vermicular movements, but did not pass away from it. It was more pointed at one end and is depicted as lying curled.

James examined 45 dogs and found six of them were infected; the animals seemed to be otherwise healthy.

Bentley,[5] in a preliminary note on this parasite, mentions that the dog in which he first found the parasites was of English breed. He describes the parasite as attacking the polymorphonuclear and transitional leucocytes. In a diagram accompanying his note, two of the parasites are shown lying in what appears to be a mononuclear leucocyte. James definitely states that the parasites were found only in the polymorphonuclear leucocytes and makes no mention of the mononuclear leucocytes.

From the above short description, it will be seen, that this parasite resembles a hæmogregarine very closely. Not long ago Laveran[6] referring to it places it in this genus. It is of extreme interest, as it invades a special leucocyte and is the first example of a mammalian *Leucocytozoön*.

Up to the time when James described this parasite our knowledge of

leucocytic parasites was confined to those described in birds by Danilewsky, Sacharoff and others, and some doubt existed as to whether the cells in which these parasites were seen were true leucocytes. Danilewsky named the parasites *Leucocytozoa*, but Laveran considered the cells in which they were contained were immature red cells and that therefore the parasites were *Hæmamœbœ*.

More recently Captain Christophers, I.M.S.,[7] has described a characteristic hæmogregarine in the blood of *Gerbillus indicus* in Madras. It presents the following appearances.

Encysted forms, each like a vermicule with a short tail sharply flexed upon the body, lie in spaces in enlarged and faintly-staining red cells. None could be identified as a young or developmental form.

Free parasites were present in blood which had been kept moist for some time. The nucleus was more elongated and was still situated near the posterior end. Some of the forms were long and slender.

In fresh preparations, the vermicules, resembling the encysted forms in appearance, exhibited active movements. They travelled rapidly, gliding along with the blunt end forward. They were also seen to twist and turn among the red cells. Some of them were observed emerging from the red cells. Only a small number kept under suitable conditions exhibited any change.

Many smears from the liver, spleen and bone-marrow were searched for developmental forms with negative results.

Christophers gives a description of a new species of *Hæmatopinus* which was found on the rats and describes the development undergone by the parasite in the body of this louse. Vermicules were seen on the fourth day in the midguts of two lice. Others removed on a later date showed large cystic bodies lying in the body cavity. Each cyst was filled with smaller oval cysts containing 6 to 8 crescentic bodies. These bodies were placed on a slide in the intestinal juice of some of the rats and it was found that most remained motionless, and some appeared to have undergone dissolution, although in the blood plasma of the rat these bodies exhibited remarkably active movements.

No further development was observed and no bodies were seen in the salivary glands or ovaries.

This mammalian hæmogregarine is confined to the red cells and is the first in which free vermicules have been described in the blood. The discovery made by Captain Christophers of its developmental cycle in the body of a louse is of extreme importance.

Still another hæmogregarine parasite has recently been described in a mammal by Dr. Andrew Balfour[8] in the blood of jerboas (*Jaculus jaculus*, or *J. gordoni* as it has been re-named,) in the Soudan. He has described three

forms of this hæmogregarine : encysted forms in the red cells, free vermicules in the plasma and a third in the liver in the form of cytocysts containing merozoits and residual protoplasm. Examination of the spleen and bone-marrow proved negative.

Laveran,[9] having seen Dr. Balfour's specimens, obtained some jerboas from Tunis belonging to the genus *Jaculus orientalis* and found they were infected with this hæmogregarine. He describes it as follows. The endoglobular elements 8μ to 8·5μ are elongated reniform bodies with a tail flexed on the body. One extremity is round and the other pointed. These bodies contain no pigment and male and female forms could not be distinguished. Free vermicules were seen in the plasma of the peripheral blood. They were more frequent in liver smears. They resembled the endoglobular parasites.

Spherical bodies measuring from 21μ to 23μ were seen in the liver smears. These cysts contained sporozoits, which were elongated with one end round and the other attenuated. The chromatin mass was well defined and large.

Laveran has named this parasite *Hæmogregarina balfouri.*

In May 1905, while searching for blood parasites in some of the smaller mammals—hedgehogs, squirrels, flying-foxes and bats, found in and around Rajkote, Kathiawar—I discovered a parasite in the large mononuclear leucocytes of the Kathiawar palm squirrel (*Sciurus palmarum* or, as it has been re-named, *Funambulus pennantii*).

A specimen was sent to Captain Christophers, who informed me that he thought the parasite was a hæmogregarine.

In all 52 squirrels were examined and 51 were found to be infected. The parasites were in large numbers in the majority and remained numerous for many weeks. It is now four months since the squirrels were first examined and the infection remains unaltered.

The parasite in fresh preparations.

On examining a drop of blood from an infected squirrel, the parasite is seen as a clear elongated worm-like body measuring 10μ in length, lying in the substance of the large mononuclear leucocytes (Fig. 1). The majority exhibit slow vermicular movements, altering their position in the cells. They may be seen lying close alongside the nucleus or at right angles to it.

The nucleus becomes compressed, and in some instances almost comes to encircle the parasite. Many of the nuclei are split into two separate parts with the parasite lying between. The invaded leucocytes show extremely slight movements. Each body is oval with one end rounder than the other. The protoplasm contains a large refractile nucleus placed about the centre and some dark dots are seen moving in the protoplasm.

The narrower end shows a distinct bend upwards simulating a tail. There is no clear space between the edge of the parasite and the substance in which it lies, and no pigment granules are seen in the protoplasm.

In fresh blood films containing a large number of parasites, some of these bodies are seen escaping from the cells. After watching one, which appears particularly active, the more rounded end will be seen to cause the cellular envelope to bulge. It ruptures and the parasite is extruded. No space is left behind, to show where the parasite was lying.

Besides the encysted forms many are seen lying free in the plasma. They have the same appearance as those in the cells (Fig. 2). The tail is most distinct when the parasite is on its side. It is quite short and ends abruptly. Most of the granules are seen between the nucleus and the tail. These forms are usually lying so that the tail is below, and is seen as a band across the protoplasm. The majority exhibit slow vermicular movements, never moving out of the field.

In films from three squirrels with swarming parasites free vermicules were seen (Fig. 3). Each is an elongated spindle-shaped body measuring 13μ to 14μ in length and from 3μ to 4μ in breadth. They were very active, performing rapid serpentine-like movements, twisting and curling about. They would approximate both ends and suddenly separate them. They attached themselves to a red cell by one end, and whirled round so fast that it was impossible to follow their movements. They very quickly glided out of sight. The nucleus was situated about the centre and consisted of a single mass. Many refractile dots were seen towards one end.

The parasite in stained films.

The parasite stains readily with Romanowsky's stain and Leishman's modification. It appears lying in the substance of the leucocyte. No distinct capsule can be made out by staining in the usual way, but by prolonged staining with Romanowsky's stain and also with Giemsa's, a faint pink outline is seen around the parasite. This under higher magnification consists of minute chromatic particles.

Each body is oval in shape with one extremity larger and rounder than the other (Fig. 4). It measures 10μ in length and 5μ across the broadest part. The protoplasm stains light-blue, somewhat darker at the two ends.

The nucleus is a large irregularly quadrilateral shaped mass, which lies about the centre. It occupies the whole of this part of the parasite encroaching on the sides. It stains deeply but not uniformly and contains three to eight vacuoles. The substance is at parts much darker than at others. Six to eleven large chromatic dots are seen in the protoplasm, more in one extremity than in the other.

The parasite is most frequently seen lying close alongside the nucleus which

adapts itself to the shape of the parasite (Fig. 4). In most of the cells no protoplasm is seen between the nucleus and the parasite. They may be also commonly seen lying between two parts of the nucleus (Fig. 5). These may be quite detached or united by a thin strand of nuclear tissue.

On three occasions two parasites were seen in one leucocyte (Fig. 6). In one cell they are lying close to each other, between the two detached parts of the nucleus. In the other cell, which was much distended, the two parasites are lying on either side of the drawn-out nucleus.

In more than one film from a heavily-infected animal, the nuclei of the parasites were seen to consist of two separate parts (Fig. 7), situated towards the extremities of the parasites. They may be quite circular or elongated. These parasites appeared in all other respects exactly similar to those described above.

The protoplasm of all the invaded leucocytes stains feebly as compared with the normal cell (Fig. 8). Even with the most intense staining with Giemsa's stain the protoplasm is indistinct and the parasite appears lying close to a nucleus.

Films were made from the blood of a Madras squirrel, when the protoplasm of the large mononuclear leucocytes was stained an intense blue (Fig. 8).

The presence of the parasite in the cell seems to cause a very definite change in the protoplasm as shown by its be haviour to stains. In one case where out of 300 large mononuclear leucocytes, 180 contained parasites, almost every one of the remaining mononuclear leucocytes consisted of a nucleus with mere traces of protoplasm.

One leucocyte showed two vacuoles lying on each side of the partially divided nucleus (Fig. 8). This would suggest that at one time these cells had harboured parasites, but that previous to the making of the film, they had left the cells. No free forms were seen close to these cells, which might have proved that the parasites were only extruded during the preparation of the film. The cells are certainly friable and break up when the film is made.

Figure 9 shows a parasite in a stained film in the act of passing out of the cell. The rounded end has already left the cell, while the more attenuated end is still in the body of the cell.

On staining the free forms with Giemsa's stain the minute structure of the parasite is clearly seen (Fig. 10). One end is globular, while the other is narrow, protoplasm consisting of a loose network, which stains blue. Scattered throughout it are minute faintly-staining pink dots, which in some places are collected together, giving a uniform pink appearance, while in others they are scattered and give the appearance of stippling.

The large chromatic dots are arranged irregularly (Fig. 11). There seems to be no definite order in their position, which might suggest any differentiation into types. The nucleus is large and stains deeply.

Some of the free forms are more attenuated than others (Fig. 13). The protoplasm stains dark-blue, and the pink stippling is marked. The nucleus, which is elongated and stains deeply, shows no vacuoles.

Figure 14 represents a vermicule from the peripheral blood stained with Romanowsky's stain. It measured 17·5μ in length and 3·5μ in breadth. The ends are attenuated, one more so than the other. The nucleus is circular and consists of a single mass without vacuoles.

Leucocyte counts of uninfected and infected squirrels.

Almost the first thing that strikes the eye on examining a blood film taken from a heavily infected animal, is the presence of a large number of mononuclear leucocytes. After making a series of counts it was thought that there was a marked increase in the number of these cells. To settle this point the blood of Madras squirrels in which the parasite was not found was examined. It is necessary to mention, that these squirrels were found to harbour the trypanosome discovered by Major Donovan, I.M.S., and described by Laveran and Mesnil.[9]

Blood counts in four cases gave the following figures. Five hundred cells were counted in each film :—

Squirrel 1.

Polymorphonuclear leucocytes	45·8
Large mononuclear leucocytes	25·
Lymphocytes	20·8
Myelocytes	5·8
Basophiles	2·4
Eosinophiles	·2

Trypanosomes few.

Squirrel 2.

Polymorphonuclear leucocytes	43·2
Large mononuclear leucocytes	32·
Lymphocytes	18·
Myelocytes	4·8
Basophiles	2·
Eosinophiles	——

No trypanosome found.

Squirrel 3.

Polymorphonuclear leucocytes	32·2
Large mononuclear leucocytes	28·2
Lymphocytes	31·6
Myelocytes	5·6
Basophiles	1·6
Eosinophiles	·8

Trypanosomes abundant.

Squirrel 4.

Polymorphonuclear leucocytes	42·6
Large mononuclear leucocytes	23·6
Lymphocytes	27·6
Myelocytes	4·8
Basophiles	1·2
Eosinophiles	·2

Trypanosomes few.

Squirrels with different degrees of infection were then examined and the figures were as follow:—

Squirrel 1, with intense infection.

Polymorphonuclear leucocytes	9·4
Large mononuclear leucocytes	80·8
Lymphocytes	9·4
Basophiles	·4
Myelocytes	.....
Eosinophiles	.....
Total number of parasites seen	326
Free parasites seen	6

Squirrel 2, with severe infection.

Polymorphonuclear leucocytes	17·8
Large mononuclear leucocytes	64·8
Lymphocytes	13·8
Myelocytes	1·6
Basophiles	1·8
Eosinophiles	·2
Total number of parasites seen	100
Free parasites seen	4

Squirrel 3, with a mild infection.

Polymorphonuclear leucocytes	33·8
Large mononuclear leucocytes	41·4
Lymphocytes	20·2
Myelocytes	2·6
Basophiles	·6
Eosinophiles	1·4
Total number of parasites seen	50
Free parasites seen	3

Squirrel 4, with mild infection.

Polymorphonuclear leucocytes	27·5
Large mononuclear leucocytes	59·5
Lymphocytes	10·5
Basophiles	·5
Myelocytes	.....
Eosinophiles	2·5
Total number of parasites seen	27
No free forms.	

It will be seen in the case of the first of these that the large mononuclears formed the bulk of the white cells, and, on comparing this figure with any of those from the Madras squirrels' counts, it will be at once apparent that there is a great increase in the numbers of this leucocyte. The Madras squirrel is taken as an example of an uninfected animal.

It was of some importance to have settled this point and, to bring it out more clearly, total counts by the field method were made from the heart-blood of the two species. The following figures were obtained :—

Madras squirrel.

	1st count.	2nd count.
Polymorphonuclear leucocytes	10,600	10,900
Large mononuclear leucocytes	4,200	4,000

Kathiawar squirrel.

	1st count.	2nd count.
Polymorphonuclear leucocytes	8,600	7,900
Large mononuclear leucocytes	10,500	11,200

This shows that we are dealing with a distinct mononuclear leucocytosis and that it varies in proportion to the degree of infection. The fact that a mammalian protozoan, which has the property of invading a special leucocyte, causes a change in the structure and an increase in the numbers of these in the peripheral blood may have some significance in relation to the origin of leucocythæmias.

In making the above counts, films were taken from the animals as near as possible about the same time of day.

The distribution of the parasite in the body.

Smears were made from the spleen, liver, kidneys, pancreas, testicles, intestinal mucosa and muscles.

The parasites were always present in considerable numbers in the spleen. They were unusually large and almost equally round at both ends (Fig. 12).

The protoplasm was blue and contained the usual chromatic dots. The nucleus in these forms was voluminous, staining less intensely and in some was worm-like in appearance. No vermicules and no young or developmental forms were seen.

Special attention was directed to the liver in view of the developmental forms found in *H. balfouri*. Fourteen squirrels were shot and smears were made from their livers. In all the parasite was found both free and in the mononuclear leucocytes, but no developmental forms. Further a heavily infected animal was killed. Thirty-four *thick* smears were made from the liver, six from the spleen and six from the kidneys. These were stained with Romanowsky's stain and were systematically examined, first with a No. 3 Leitz, then with a $\frac{1}{12}$ oil immersion. No cyst or other form suggestive of development were seen. The parasites were numerous, both free and encysted, and had the appearances of those in the spleen (Fig. 12). The parasites were scarce in the other tissues.

The majority of the Kathiawar squirrels are infected with a worm in the abdomen which is seen free and encysted in the abdominal muscles and diaphragm. No filarial embryos were seen in the blood.

The parasite in the body of the squirrel louse.

This louse has been identified for me by Professor Neumann and Dr. Newstead as an undescribed species of *Hæmatopinus*.

It is found in large numbers among the long hairs of the squirrel. It clings tenaciously to the hairs and is comparatively slow in its movements. The ova measure ·6 m.m. and are laid on the hairs of the legs and tail.

Male.—When full-grown is ·9 m.m. in length, the head is triangular in shape with two antennæ springing from the upper angles. Each consists of five segments. The third is most attenuated and the basal most globular. A large spine springs from the upper surface of this segment. The eyes are small and are situated behind the last antennal joint.

One short and one longer spine spring from the dorsum of the head.

The junction of the head and thorax is marked by a constriction. The thorax consists of a large dorsal chitinous plate. It has one spine opposite the first leg and two opposite the second. The legs, three on each side, consist of four segments. The third leg is the most developed. The basal segment is large and has a spine on the under-surface. The third segment is smaller and not quite so globular. It has a few spines on it. The second is larger and has two or three marked spines. The first segment is globular and has the claw attached to it by a triangular joint. Below the claw there is a large pad. The size of the various segments of the legs increases from before backwards: this is most marked in the case of the claws.

The abdomen consists of nine segments. On the dorsum of each segment there is a fringe of hairs and on each side there is a group of four or five. The last segment carries one very long hair on each side. This segment ends in a conical-shaped process from which the penis, a bifid structure consisting of two long chitinous rods, protrudes.

Female.—The head of the female is smaller than that of the male; the third antennal segment is not attenuated as in the male, and there is a marked spine on the basal segment.

The abdomen is made up of nine segments and is about twice as long as in the male. The dorsal surface has two chitinous bands for each segment. The last segment ends in a flat surface and has a fringe of six hairs placed laterally. On the ventral aspect the hairs are arranged as in the male, except that the first segment has four, the seventh has two and the eighth three. The legs are the same as in the male.

Each female lays on an average five eggs. The young hatch out into fully developed lice after three to four days.

This louse is closely related to *H. antennatus*, Osborn, and *H. spinniger*, Denny.

On one occasion a small acarid was found parasitic on a louse, clinging to the abdomen.

Lice were the only octoparasites that were seen on the squirrels and at Captain Christophers' suggestion I decided to try to ascertain whether developmental changes are undergone by the parasite in this louse.

A young male squirrel with swarming parasites was selected and 42 lice were placed on it from other squirrels. On the second day two lice were removed and examined. The midguts were dissected out and searched in the fresh condition. Active vermicules were seen exactly similar to those in the peripheral blood. On staining one of these, it was found to measure 15·5μ, was spindle-shaped with one extremity slender and more pointed (Fig. 15). The protoplasm stained blue throughout and had a stippled appearance due to minute chromatic dots. The nucleus was situated about the centre, and consisted of a circular mass of protoplasm which stained uniformly dark. Between it and the thin end were six large chromatic dots. No other form was seen in the midgut.

On the fourth day two more lice were examined, but besides the vermicules nothing else was seen which might have suggested further development. On the fifth day an adult female louse was removed. The midgut was dissected out without being ruptured. It contained active vermicules and encysted parasites as the blood was fresh. The remainder of the louse was carefully examined and one vermicule was seen slowly moving about in the body cavity. On staining it was found to measure 18μ with one end attenuated and

slightly bent, the other was broader (Fig. 16). The protoplasm stained blue, darker towards the pointed end. Two very distinct vacuoles were seen, one at the extreme end and the other deeper and to the side of the broader end. Seven large chromatic dots were seen near the vacuoles. The nucleus consisted of a collection of chromatic rods arranged irregularly.

Lice were examined regularly for ten days after being placed on the squirrel, but, with the exception of vermicules, nothing suggesting any further development was seen either in the gut or in the body cavity. No parasitic bodies were seen in the salivary glands or ovaries.

Twenty-four lice were placed on a Madras squirrel from which all the lice that could be found were removed. This squirrel was kept separate but up to the twelfth day the peripheral blood remained free from infection.

Conclusions.

It will be seen that the description of this parasite, agrees in every detail with that of a hæmogregarine. The parasite is highly specialised, in that it selects the large mononuclear leucocyte for its host. Though closely related to *Leucocytozoön canis* (James), it differs in not having a cytocyst and in possessing a tail. I therefore propose provisionally naming it *Leucocytozoön funambuli*.

As in the case of *Hæmogregarina gerbilli* (Christophers), no developmental forms were found in the organs and the infection remains unaltered for long periods. The squirrels, though harbouring two parasitic worms, were to all appearances as active as the Madras species. It is particularly interesting to note, that trypanosomes were never found in the blood of the Kathiawar squirrel, which seems to point to a localised distribution of the blood parasites in very closely related mammals.

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FIGURE 1.—Parasites as seen in the large mononuclear leucocytes in fresh preparations of the peripheral blood.

FIGURE 2.—Free parasites seen in the fresh preparations of the peripheral blood. The tail is shown in one lying on its side.

FIGURE 3.—Vermicule seen in fresh preparation of peripheral blood.

FIGURE 4.—Parasite seen in the large mononuclear lying alongside the nucleus, stained with Romanowsky's stain. Two polymorphonuclear leucocytes are also shown.

FIGURE 5.—Parasite stained with Romanowsky's stain lying in between the divided nucleus.

FIGURE 6.—Two mononuclears stained with Romanowsky's stain showing the presence of two parasites in each.

FIGURE 7.—Parasites stained with Romanowsky's stain showing the nucleus in two separate parts.

FIGURE 8.—On the left is a mononuclear leucocyte showing the faintly-staining protoplasm with two vacuoles. On the right the normal mononuclear leucocyte from the Madras squirrel showing the appearance when stained with Romanowsky's stain.

FIGURE 9.—Parasite escaping from a cell stained with Romanowsky's stain.

FIGURE 10.—Free parasite stained with Giemsa's stain showing minute structure.

FIGURE 11.—Three free forms stained with Giemsa's stain showing the arrangement of the large chromatic particles.

FIGURE 12.—Four free parasites seen in spleen smears stained with Romanowsky's stain showing the voluminous nucleus and also its worm-like appearance seen in smears.

FIGURE 13.—Free attenuated form of parasite in peripheral blood stained with Romanowsky's stain showing the elongated nucleus.

FIGURE 14.—Vermicule from peripheral blood stained with Romanowsky's stain.

FIGURE 15.—Vermicule from the mid-gut of the squirrel louse, stained with Romanowsky's stain.

FIGURE 16.—Large vermicule seen in the body cavity of the louse stained with Romanowsky's stain.

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Fig 1

Fig 2

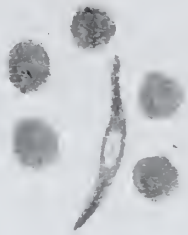

Fig. 3.

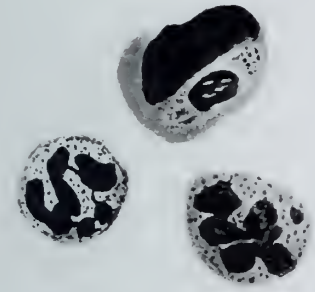

Fig. 4

Fig 5

Fig 6

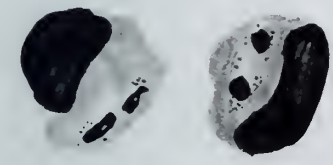

Fig. 7

Fig 8

Fig 9

Fig 10.

Fig. 11

Fig. 12

Fig. 13

Fig. 14.

Fig. 15.

Fig. 16

Photogravure.

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BY

CAPTAIN W. S. PATTON, M.B., I.M.S.

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